



The Biokinetic Spectrum for Temperature and optimal Darwinian fitness

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ABSTRACT

Darwinian fitness is maximised at a temperature below T_{opt} , but what this temperature is remains unclear. By linking our previous work on the Biokinetic Spectrum for Temperature with a model for temperature-dependent biological growth rate we obtain a plausible value for such a temperature. We find this approach reveals considerable commonalities in how life responds to temperature with implications that follow in evolution, physiology and ecology.

We described a data set consisting of 17,021 observations of temperature-dependent population growth rates from 2411 bacterial, archaeal and eukaryal strains. We fitted a thermodynamic model to describe the strains' temperature-dependent growth rate curves that assumed growth was limited by a single rate-limiting enzyme. We defined U_{mes} as an empirical measure of the temperature at which strains grew as fast and also as efficiently as possible. We propose that Darwinian fitness is optimised at U_{mes} by trading-off growth rate and physiological efficiency.

Using the full data set we calculated the Biokinetic Spectrum for Temperature (BKST): the distribution of temperature-dependent growth rates for each temperature. We used quantile regression to fit alternative models to the BKST to obtain quantile curves. A quantile is a value that contains a particular proportion of the data. The quantile curves suggested commonalities in temperature-dependencies spanning taxa and ecotype, consistent with the single rate-limiting enzyme concept. We showed that on the log scale, the slopes of the quantile curves were the same as the slopes of the thermodynamic model growth curves at U_{mes} . This was true for Bacteria, Archaea, and Eukarya, and across other conditions (pH, water activity, metabolic type and trophic type). We showed that the quantile curves were the loci of the temperatures and growth rates that optimised Darwinian fitness for each strain at a given temperature-dependence and independently of other conditions.

The quantile curves for Archaea and Bacteria shared a number of similarities attributable to the influence of the properties of water on protein folding. Other implications have impact on evolutionary biology, ecology, and physiology. The model predicts the existence of eurythermic strains that grow with about equal efficiency over a broad temperature range. These strains will have higher evolutionary rates with lower mutational costs that are independent of environmental conditions, a factor likely to have been significant during the Precambrian if the early Earth was warmer than today. The model predicts that random mutations are likely to result in shifts along the quantile curves and not across them. It predicts that some psychrophiles will be capable of performing well under climate change, and that selection will favour faster growth rates as the temperature increases. Last, it predicts trade-offs between growth rate and soma production, so that temperature-dependence, and possibly Darwinian fitness, remain constant over a broad temperature range and growth rates.

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1. Introduction

A species only grows within the temperature range to which it is adapted. It has a maximum growth rate and a maximum

temperature-dependence of growth rate. Considering the temperature range, growth has been recorded from well below the freezing point of pure water up to an observed maximum of 122 °C (Takai et al., 2008). Growth rates are limited, with the maximum known that of *Clostridium perfringens* with a doubling time of 6.3 min (Labbe and Huang, 1995). However, the temperature dependence of growth rate is more contentious with some claiming that, after adjustment for organism size, it is invari-

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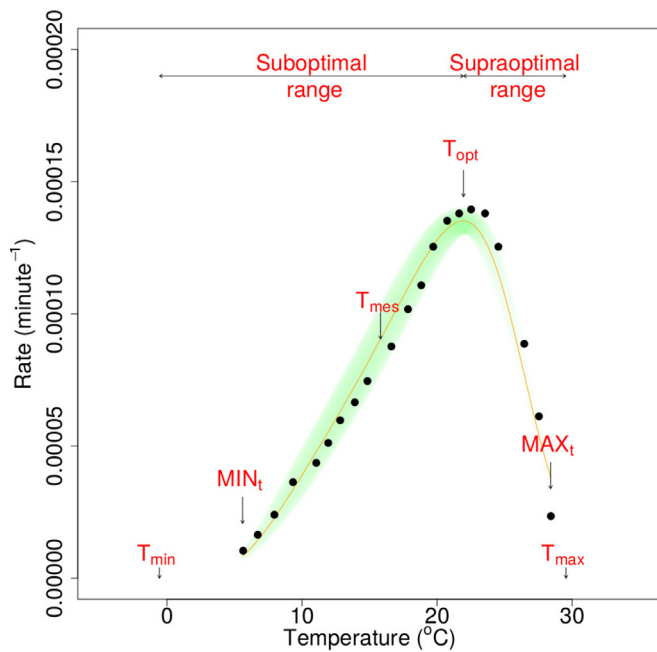


Fig. 1. Example of growth rate data for a single microbial strain. The black circles (•) are observed temperature-dependent growth rates for *Methanococcoides burtonii*. Shown are observed minimum and maximum temperatures for growth MIN_t and MAX_t , temperature of maximum growth rate T_{opt} , temperature of maximum enzyme stability T_{mes} , and predicted minimum and maximum temperatures for growth T_{min} and T_{max} . The T_{min} and T_{max} are obtained using the square root model from Ratkowsky et al. (1983). The solid line is the fitted growth curve obtained using the thermodynamic model (q.v., Methods). The shaded region schematically shows the enzyme stability from the thermodynamic model. The width of the shaded region is proportional to the degree of enzyme stability (not to scale).

ant (Brown et al., 2004), and others that it is variable (Dell et al., 2011), presumably with upper and lower limits.

Population growth rates can be quantified by specific or intrinsic growth rates (Birch, 1948; McMeekin et al., 1993). In this paper we refer to these as growth rates and term plots of growth rates versus temperature as growth curves. The temperature at which the maximum growth rate is reached is T_{opt} , dividing the growth curve into suboptimal and supraoptimal regions, after which growth usually declines more rapidly (Fig. 1). Growth curves are ultimately determined by the rate of chemical reactions within the cell (Stegelmann et al., 2009). These chemical processes involve enzymes that denature at sufficiently low or high temperatures (Somero, 1995). It has been proposed that temperature-dependent population growth rates are controlled by a single rate-limiting, enzyme-catalyzed reaction within the cell (e.g., Ratkowsky et al., 2005). We refer to this putative mechanism as the Master Reaction System (MRS). A thermodynamic-based model for temperature-dependent cell growth rate has been developed based on the MRS hypothesis (Corkrey et al., 2012; Ratkowsky et al., 2005). While the thermodynamic model has been very successful, it has some limitations. First we note the positive aspects and then the limitations.

The thermodynamic model has successfully and accurately modelled the relative growth rates of strains in all three Domains of life (Bacteria, Archaea, Eukarya) ranging from psychrophilic to hyperthermophilic and spanning temperatures from -10 °C up to 122 °C (Corkrey et al., 2012), obtaining values of thermodynamic parameters that, for the most part, were consistent with expectations from biochemistry (Corkrey et al., 2014). It provided excellent fits to absolute growth rates of 230 strains of unicellular and multicellular organisms (Corkrey et al., 2014). However, the thermodynamic model provides no information on the range of growth rates

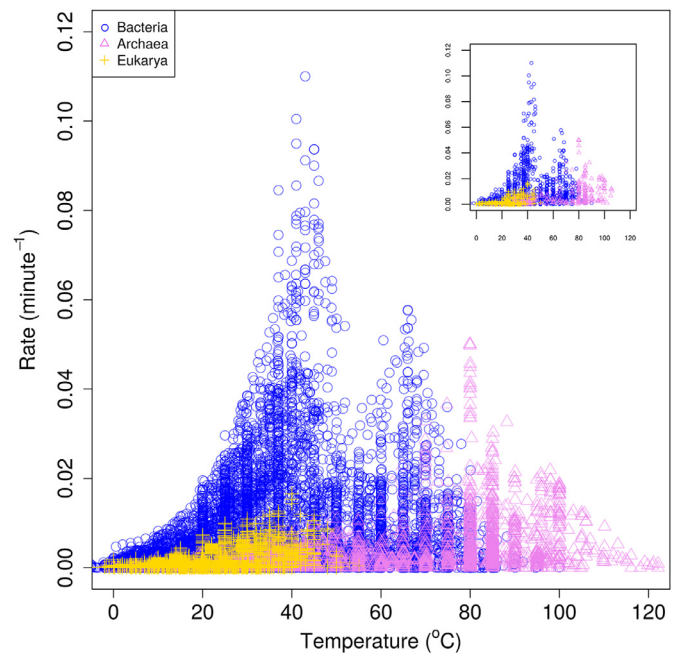


Fig. 2. The BKST showing all three domains. Shown are 17021 growth rates versus temperature from the three domains (q.v., Methods). The blue circles are for Bacteria; violet triangles for Archaea; and gold crosses for Eukarya. The inset shows the BKST plotted using a single datum per strain. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

or growth rate dependencies that may be possible. Such information would be of use in astrobiology, for example, where the limits of life are of current concern (Horneck et al., 2016).

The Biokinetic Spectrum for Temperature (BKST) has been recently described by us (Corkrey et al., 2016). It consists of the distribution of temperature-dependent growth rates against environmental temperature (Fig. 2). The BKST included the fastest growing organisms known at each temperature so that the observed upper limit was considered to represent the maximum growth rate at a given temperature for any life on Earth. The upper boundaries of the BKST were observed to follow exponential curves, below which were strains growing at varying, but slower rates. The BKST represents a significant, and perhaps universal, biological phenomenon that will prove insightful of the influence temperature has on life.

The sharpness of the upper boundary of the BKST suggested it may provide information on the limits imposed by evolution on growth rates. We hoped it would illuminate why organisms have particular growth rates and how they related to growth rate dependencies. We were also interested in knowing if growth rates could be suitable proxies for Darwinian fitness (Martin and Huey, 2008). To this end, in this paper we fit the thermodynamic model to growth curves to obtain measures of growth rate dependencies and the temperature ranges of protein denaturation, link these measurements to the BKST, and interpret its structure using the concept of Darwinian fitness.

2. Methods

2.1. Data

Some of the data used have been previously published (Corkrey et al., 2012) and the remainder obtained by searching the peer-reviewed literature. Details on our search methods have been published elsewhere (Corkrey et al., 2016). We located studies that reported biological growth rates and

their dependence on temperature, pH and water activity (a_w) or salinity. Some sources reported salinity but did not report water activity. For these we estimated water activities from the NaCl concentration using the predictions compiled by [Resnik and Chirife \(1988\)](#), but omitted cases where the NaCl concentrations were greater than 26%. We prefer the use of the word strain in this paper rather than species because some data sets refer to a single species grown under different conditions, or the same species grown by different researchers. We scaled all the growth rates to the same units: growth per minute versus temperature in Celsius. The literature sources for the data are given in Table S1.

2.2. The Thermodynamic Model

We used a previously described model ([Corkrey et al., 2014; 2012; Ratkowsky et al., 2005; Ross, 1997](#)) to fit growth curves to the temperature-dependent population growth rate data. We show the central equation of the model in [Eq. \(1\)](#). This model assumed that the temperature-dependent growth rate of an organism was limited under all conditions by a single rate-limiting Master Reaction System (MRS). In the thermodynamic model the observed growth rate is denoted by r and the modelled growth rate by F . The numerator, $(T \cdot \exp(c - \Delta H_A^\ddagger / RT))$, can be considered to be an Arrhenius function that is often used to describe the rate of chemical reactions. As the temperature rises sufficiently high, or drops low enough, the enzyme is more likely to denature. This will have the effect of reducing growth since a greater proportion of the enzyme in the cell will be denatured. The combination of the numerator and denominator is therefore intended to describe the reaction rate of the enzyme as the temperature changes. Since the enzyme is assumed to control the organism growth rate, the thermodynamic model essentially describes the growth rate of the organism.

$$F = \frac{T \cdot \exp\left(c - \frac{\Delta H_A^\ddagger}{RT}\right)}{1 + \exp\left(-n \cdot \frac{\Delta H^* - T \cdot \Delta S^* + \Delta C_p \cdot (T - T_H^* - T \cdot \ln\left(\frac{T}{T_S^*}\right))}{RT}\right)} \quad (1)$$

In [Eq. \(1\)](#) $\ln$ is the natural logarithm function; R is the gas constant (8.314 J/K mol); c is a scaling constant; $\Delta H_A^\ddagger$ is the enthalpy of activation (J/mol); T is the temperature (K); ΔC_p is the heat capacity change (J/K mol-amino acid residue) upon denaturation of the rate-limiting reaction; n is the number of amino acid residues of the putative rate-controlling enzyme; ΔH^* is the enthalpy change (J/mol amino acid residue) at T_H^* , the convergence temperature for enthalpy (K) of protein unfolding; ΔS^* is the entropy change (J/K) at T_S^* , the convergence temperature for entropy (K) of protein unfolding. The notation follows that of [Murphy et al. \(1990\)](#) except that ΔH^* , ΔS^* , and ΔC_p are expressed per mole of amino acid residue and we introduce the n parameter ([Ratkowsky et al., 2005](#)). The parameters, ΔH^* and ΔS^* , which are discussed elsewhere with a change in notation (i.e. ΔH^u and ΔS^u) ([Makhatadze and Privalov, 1993; Privalov and Makhatadze, 1993](#)), arise in the context of enthalpy-entropy compensation ([Privalov and Gill, 1988](#)). We follow [Baldwin \(1986\)](#) in assuming ΔH^* and ΔS^* to be constants for hydrocarbons and also [Privalov and Gill \(1988\)](#) in assuming T_H^* and T_S^* are universal properties of proteins, although they use a different notation.

While there are eight parameters, only four (c , $\Delta H_A^\ddagger$, ΔC_p , n), are used to fit individual strains, the other four (ΔH^* , T_H^* , ΔS^* , T_S^*) are assumed to be common to all strains. We have found in practice that to obtain excellent fits it is mandatory that all parameters remain distinct. For example, the global parameters (ΔH^* , T_H^* , ΔS^* , T_S^*) need to be regarded as four free parameters. Similarly, the parameters (c , $\Delta H_A^\ddagger$, ΔC_p , n) must be separately estimated for each strain.

We fitted the thermodynamic model using Bayesian methods to allow for uncertainties in parameters and data to be incorporated in a natural way through the appropriate prior specification. We controlled homogeneity of variance by modelling the square root of the growth rate. We assumed that the square root of the observed growth rate had a Gaussian distribution with a mean given by the square root of the modelled value and had an unknown precision (reciprocal variance),

$$\sqrt{r} \sim N(\sqrt{F}, \psi).$$

Since this was a Bayesian model it was necessary to assign prior probability distributions to the parameters that reflected our belief in their values before fitting the model. For c , $\Delta H_A^\ddagger$ and n we assigned normal prior distributions to the strain parameters. The means of the strain parameters were specific to a thermal group which were defined based on quantiles of the strains' T_{opt} . A quantile is a value that contains a particular proportion of the data. The prior means were assigned uniform distributions and their precisions were assigned gamma distributions. In the case of ΔC_p we used a simple prior since this parameter was always well supported by data. In summary, the prior specifications for strain j in thermal group d were

$$\begin{aligned} c_j &\sim N(\gamma_d, \tau_d), \\ \gamma_d &\sim \text{Unif}(-20, 150), \quad \tau_d \sim G(10^{-3}, 10^{-3}), \\ \Delta H_{A_j}^\ddagger &\sim N(\eta_d, \frac{\theta_d}{10^8}), \\ \eta_d &\sim \text{Unif}(0.01, 500000), \quad \theta_d \sim G(10^{-3}, 10^{-3}), \\ \Delta C_{p_j} &\sim N(65, 0.0001), \\ n_j &\sim N(\lambda_d, \frac{\tau_d}{10^6}), \\ \lambda_d &\sim \text{Unif}(1, 20000), \quad \tau_d \sim G(10^{-3}, 10^{-3}). \end{aligned}$$

The priors for the remaining parameters were

$$\begin{aligned} \Delta H^* &\sim \text{Unif}(3000, 7000), \quad T_H^* \sim \text{Unif}(320, 420), \\ \Delta S^* &\sim \text{Unif}(10, 30), \quad T_S^* \sim \text{Unif}(320, 420), \\ \psi &\sim G(0.001, 0.001). \end{aligned}$$

Inference was obtained in the form of posterior means, variances and credible intervals using Markov chain Monte Carlo (MCMC) simulation ([Brooks, 1998](#)). A credible interval contains a parameter with specified probability.

We chose to update the parameters of each strain and thermal group means using Haario updates ([Haario et al., 2001](#)). For the universal parameters we used adaptive direction sampling ([Gilks et al., 1994](#)) combined with a low probability stepping-stone proposal ([Gilks and Roberts, 1996](#)). The models were run for 5,000,000 iterations and the last 50% of iterations retained for further analysis. The model was fitted to the entire growth curve of the strains including both suboptimal and supraoptimal regions and for those strains that had 8 or more observations, but excluded those that lacked a well-defined peak at T_{opt} . Since the bulk of the data were from microbes size-correction was not of interest. Any effect of size was absorbed into the c parameter.

As noted above, the reciprocal of the denominator of [Eq. \(1\)](#) gives the probability of the native state ([Ratkowsky et al., 2005](#)), which is

$$P_{NS} = \frac{1}{1 + \exp\left(-n \cdot \frac{\Delta H^* - T \cdot \Delta S^* + \Delta C_p \cdot (T - T_H^* - T \cdot \ln\left(\frac{T}{T_S^*}\right))}{RT}\right)}. \quad (2)$$

Using [Eq. \(2\)](#) we plotted the P_{NS} curve versus temperature. The peak of the P_{NS} curve occurs at the temperature T_{mes} , the temperature at which denaturation is minimized, given by

$$T_{mes} = T_H^* - \frac{\Delta H^*}{\Delta C_p}.$$

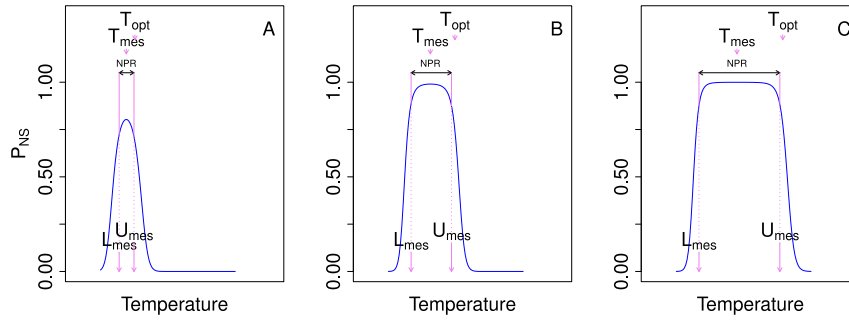


Fig. 3. Examples of protein native stability curves. Shown are protein native stabilities (P_{NS}) for three arbitrary strains illustrating the diversity in forms the curve can display. The curves reach a maximum at T_{mes} , the temperature at which the enzyme is most likely to be stable. We also show the normal physiological range (NPR) centred around T_{mes} which is bounded by (L_{mes} , U_{mes}). By way of comparison, we also include the T_{opt} for each strain.

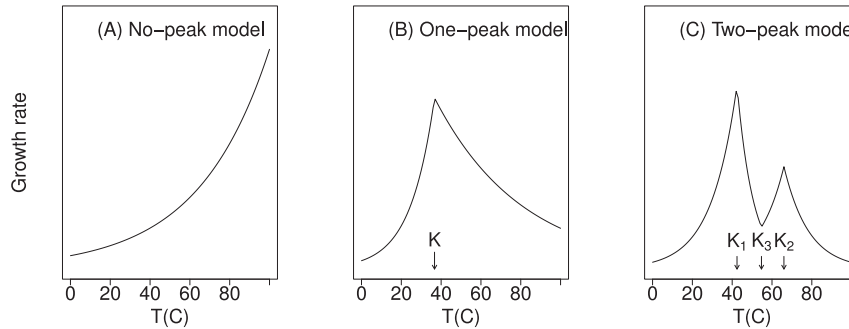


Fig. 4. Schematic of the three alternative BKS models used in the quantile regressions. For the one-peak model K is the temperature of the peak. For the two-peak model K_1 and K_2 are the temperatures of the two peaks, and K_3 is the temperature of the minimum of the trough between the peaks.

Growth rate reaches a maximum at T_{opt} , but the thermodynamic model assumes that the organism's metabolism operates most 'efficiently' at T_{mes} . The P_{NS} curve has usually been found to be peaked but could also be flat-topped (Corkrey et al., 2012; Ratkowsky et al., 2005). We denoted the range of temperatures centred on T_{mes} where the enzyme was most likely to be stable as the normal physiological range (NPR). For the peaked curves this was exactly at T_{mes} whereas for the flat-topped curves it was a range of temperatures centred on T_{mes} . In strains with a flat-topped P_{NS} curve the cell could operate with almost equal efficiency over the NPR. To quantify the NPR we defined it to be the range of temperatures centred around T_{mes} where $P > 0.9 \cdot \max(P_{NS})$; i.e. 90% of the maximum height of the P_{NS} curve. We defined the temperatures at the lower and upper ends of the NPR as L_{mes} and U_{mes} , respectively. We illustrate these quantities in Fig. 3.

2.3. The Biokinetic Spectrum for Temperature

We used Bayesian quantile regression to describe the BKST (Fig. 2). Quantile regression is a technique used to estimate quantiles of the response variable given values of a predictor. A quantile is a value that contains a particular proportion of the data. In our previous description of the BKST (Corkrey et al., 2016) we only used a model for the quantiles that had a single peak since the aim in that study was to describe simply the general properties of the spectrum. The one-peak model had the same basic form as the BKST. However, we were also familiar with a trough that appears in the spectrum at around 50 °C (see Fig. 2). It was unclear if the trough was a real phenomenon or a result of insufficient data at that temperature. Therefore we decided in this study to allow for the trough by including a two-peak model. This model possessed a trough located between the two peaks. Purely for reasons of symmetry we also included a no-peak model, so that, in total, we would have three models with zero, one, or two peaks. The

overall aim was to facilitate a closer examination of the BKST using three alternative models, one of which lacked a peak, one that had a single peak, and the last that had two peaks. The definitions of the BKST models were: no-peak model:

$$r = \exp(A + B \cdot T,) \quad (3a)$$

condition: $B > 0$;

one-peak model:

$$r = \begin{cases} \exp(A + B \cdot T), & \text{if } T \leq K, \\ \exp(C + D \cdot T), & \text{if } T > K, \end{cases} \quad (3b)$$

where $C = A - (D - B) \cdot K$,

condition: $B > 0$, $D < 0$;

two-peak model:

$$r = \begin{cases} \exp(A_1 + B_1 \cdot T), & \text{if } T \leq K_1, \\ \exp(C_1 + D_1 \cdot T), & \text{if } T > K_1 \wedge T \leq K_3, \\ \exp(A_2 + B_2 \cdot T), & \text{if } T > K_3 \wedge T \leq K_2, \\ \exp(C_2 + D_2 \cdot T), & \text{if } T > K_2, \end{cases} \quad (3c)$$

where $C_1 = A_1 - (D_1 - B_1) \cdot K_1$,

$C_2 = A_2 - (D_2 - B_2) \cdot K_2$, and

$K_3 = (A_2 - C_1)/(D_1 - B_2)$

condition: $B_1 > 0$, $B_2 > 0$, $D_1 < 0$, $D_2 < 0$;

where r is the predicted rate; T is the temperature; A , B , K , D , A_1 , B_1 , K_1 , D_1 , A_2 , B_2 , K_2 , and D_2 are stochastic parameters; and C , C_1 , C_2 , and K_3 are calculated from the other parameters. The three alternative BKST models are schematically illustrated in Fig. 4. The no-peak model in Eq. (3a) had two parameters and corresponded to a simple exponential curve. The one-peak model in Eq. (3b) had four parameters and consisted of two back-to-back exponential curves with a peak at the temperature labelled K . The two-peak model in Eq. (3c) had eight parameters and described four exponential curves forming two peaks that occurred

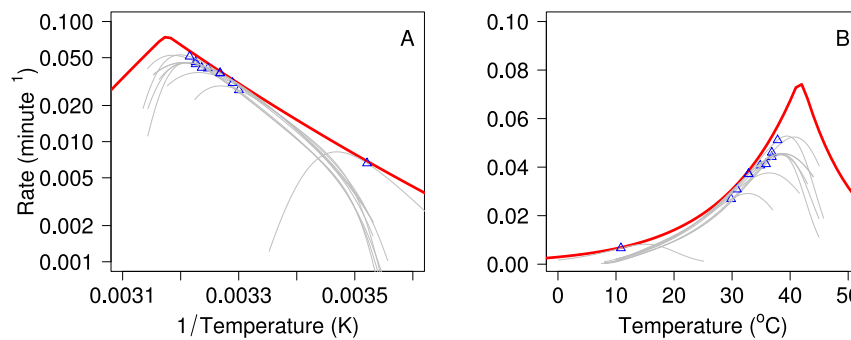


Fig. 5. Schematic of the intersections of growth curves with a quantile curve. Shown are examples of growth curves tangential to a quantile curve. The locations of U_{mes} for each strain are shown as triangles. (A): logarithm of the growth rate versus reciprocal absolute temperature. (B): growth rate versus temperature.

at the temperatures labelled K_1 and K_2 and with a trough between them at the temperature labelled K_3 . The B and D parameters in Eqs. (3b) and (3c) controlled the slope and curvature of the quantile curves while A and C controlled the heights of the curves at 0 °C. These functions were calculated separately for quantiles (1%, 3%, 5%, ..., 99%). We refer to the fits for the quantiles as quantile curves. Quantile curves were calculated by domain (Bacteria, Archaea and unicellular Eukarya) as well as by subgroups of Bacteria: tertiles of pH and a_w , metabolic type (aerobes, anaerobes, and facultative anaerobes), and trophic type (heterotrophs and autotrophs). Tertiles are the values that divide a sample into three equal parts. Multicellular Eukarya were excluded to maintain comparability with the other domains and due to the small sample size involved. We chose to work within a Bayesian framework for consistency with the thermodynamic model. The stochastic parameters were assigned normal distribution priors with fixed means and precisions. The priors for each BKS model were:

no-peak model:

$$A \sim N(-5.4, 0.04), \quad B \sim N(0.08, 1.0);$$

one-peak model:

$$A_1 \sim N(-5.4, 0.04), \quad B_1 \sim N(0.08, 1.0), \\ \ln K_1 \sim N(4.78, 0.0001), \quad D_1 \sim N(-10^{-8}, 10^2);$$

two-peak model:

$$A_1 \sim N(-5.4, 0.04), \quad B_1 \sim N(0.08, 1.0), \\ \ln K_1 \sim N(4.78, 0.0001), \quad D_1 \sim N(-10^{-8}, 10^2), \\ A_2 \sim N(-5.4, 0.04), \quad B_2 \sim N(0.08, 1.0), \\ \ln K_2 \sim N(4.78, 0.0001), \quad D_2 \sim N(-10^{-8}, 10^2)$$

where B , B_1 , B_2 were restricted to positive values and D , D_1 , D_2 to negative values. Inference was obtained in the form of posterior means and variances using MCMC. The details of the fitting procedure are given elsewhere (Yu and Moyeed, 2001). The models were run for 1,000,000 iterations and the last 50% of iterations retained for further analysis. The choice of model was evaluated separately for each quantile by comparing deviance information criteria (DIC), calculated as $DIC = -2 \cdot E(Lk) + 0.5 \cdot Var(2 \cdot Lk)$, where Lk was the likelihood at each iteration (Gelman et al., 2014), after smoothing using a running-median, E is the expectation, and Var is the variance. Prediction accuracies were assessed per quantile by comparing the difference of the mean predicted quantile to the observed quantile of the data at each temperature.

For each strain we used recursive partitioning (Zhang and Singer, 2010) to investigate the association of taxonomic, metabolic and trophic variables with quantiles. Recursive partitioning used the *ctree* function in R, version 3.4 (Hothorn et al., 2006). The method splits the outcome variable in an optimal way using the explanatory variables. The explanatory variables were kingdom, phylum, class, order, family, aerobic status, and trophic type.

2.4. Slopes and slope differences of quantile and growth curves

We assumed that the form of the BKST was the result of the various growth curves of the organisms within it. Therefore, we surmised the properties of the BKST should be related to the shapes of the growth curves. Specifically, the shapes of the quantile curves represent the change in performances for species as temperature increased. This change in the shape of growth curves should determine the form of the quantile curves. To assess this we needed to find which part of the growth curves changed most closely in accordance with the quantile curves. Due to their geometry, there could only be a single point on the growth curve at which it was parallel to an intersecting quantile curve. To locate this point, we compared the differences in slopes of quantile and growth curves, which would be zero at the point where they were parallel. This would be the point on the growth curves that determined the quantile curves, and hence the shape of the BKST. We were also interested if the contact point was the same for all species or whether it differed.

Logarithms of the quantile curves become straight sloping lines. The logarithms of the growth curves are also relatively linear within most of the strain's suboptimal range, beyond which they depart non-linearly. We refer to the slopes of the logarithm-transformed quantile or growth curves as log-slopes.

We identified which growth curves intersected but did not exceed each ascending quantile curve. The growth curves of interest were those that just touched the quantile curve. At the single point of contact the quantile and growth curves were necessarily parallel; i.e., they had the same log-slope. In the ideal, this would be the temperature at which the quantile and growth curves touched (i.e., the osculation). If a growth curve exceeded in height that of the quantile curve then there would have to be another lower quantile curve that just touched the growth curve. In practice, curves were only fitted for the quantiles 1%, 3%, 5%, ..., 99%, so we identified the lowest of these quantile curves that equalled or exceeded the growth curve. We then determined the difference in log-slopes at temperatures within 20 °C above and below U_{mes} . We illustrate this geometry in Fig. 5. We calculated these log-slope differences for each domain and subgroups of Bacteria (metabolic type, tertiles of pH and a_w , and trophic type), there being insufficient data to examine subgroups of Archaea and Eukarya.

3. Results

We summarise the sample used to construct the BKST in Fig. 1. The full collated data set and data sources are given in the Supplementary Materials. The data used to fit the thermodynamic model consisted of 389 strains and 5441 observations. The posterior parameter estimates are given in Table S2 and fitted curves for all strains are shown in Figs. S1 to S25.

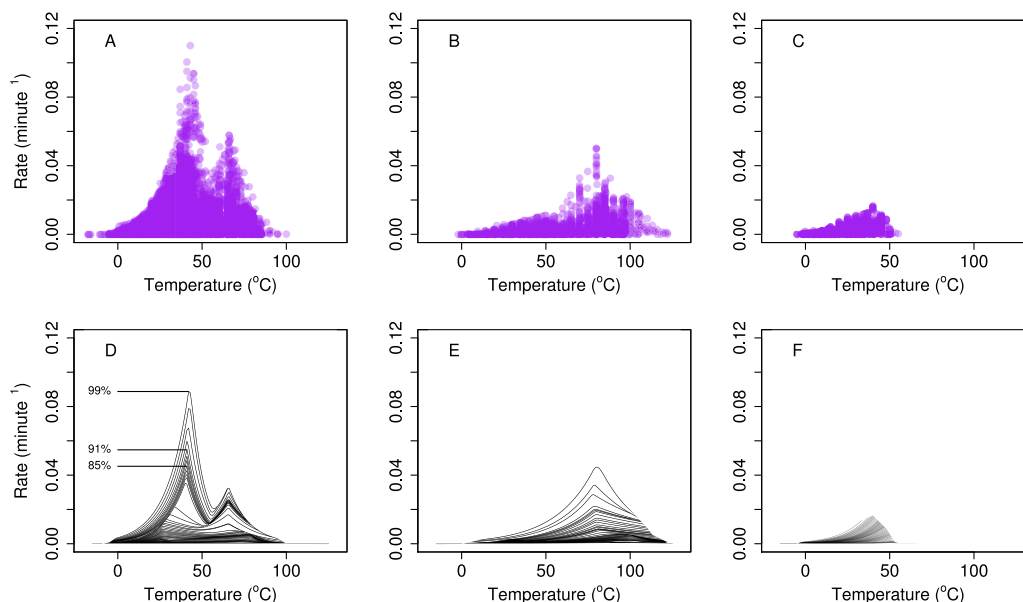


Fig. 6. The BKST with fitted quantile curves by domain. (A, D): Bacteria. (B, E): Archaea. (C, F): unicellular Eukarya. Shown in the upper row are observed growth rate data. In the lower row are the fitted curves for quantiles (1%, 3%, 5%, ..., 99%). Selected quantile curves are labelled for 3 quantiles in panel D. For example, 85% of the observed data are expected to be below the 85% quantile curve.

Table 1
Sample characteristics of data used to construct the BKST.

Property	Value
Number of observations	17,021
Number of strains	2411
Number of strains represented by a single datum	406
Median number of observations per strain	6
Number of bacterial phyla	28
Number of archaeal phyla	4
Number of eukaryal phyla	20
Number of bacterial strains	1381
Number of archaeal strains	458
Number of eukaryal strains	572
Number of multicellular strains	194
Bacteria, pH tertile ranges	0.9–6.5, 6.5–7.2, 7.2–13.0
Bacteria, a_w tertile ranges	0.8–0.98, 0.98–0.996, 0.996–1.0
Bacteria, metabolic subgroups (n)	422 aerobes
	525 anaerobes
	390 facultative anaerobes
Bacteria, trophic subgroups (n)	1142 heterotrophs
	204 autotrophs

3.1. The Biokinetic Spectrum for Temperature

The BKST of the complete data set is shown in Fig. 2. The BKST did not change much if only a single datum per strain was plotted, except that its form became less clearly defined (see the inset to Fig. 2). The higher peak at 43 °C was dominated by Bacteria, also present in a second peak at 66 °C. The fastest growing Archaea achieved a rate less than half the rate of the fastest Bacteria (0.050 min.^{−1} vs. 0.110 min.^{−1}). In comparison, Eukarya, at their fastest only achieved 0.016 min.^{−1}, and were limited to temperature below 55.1 °C.

We calculated quantile curves separately by domain (Fig. 6). Bacteria (Fig. 6(A,D)) were best described by the two-peak model, Archaea (Fig. 6(B,E)) by the one-peak model for the upper quantiles and the two-peak model for the lower quantiles (though this is difficult to discern in Fig. 6(E)), and Eukarya (Fig. 6(C,F)) by the no-peak model for all quantiles. The prediction accuracies were below

8% for Bacteria, 13% for Bacteria, 14% for unicellular Eukarya. The Bacteria showed a discontinuity in the quantile curves where there was an abrupt change in spacing. There were two peaks seen in Bacteria at 43.7 °C and 65.8 °C, a single peak in Archaea at 78.4 °C, and, as noted, there was no peak in Eukarya.

BKST substructure. By plotting the logarithm of growth rate versus reciprocal absolute temperature the exponential curves became very slightly curved lines (Fig. 7). Bacteria (Fig. 7(A,D)) displayed the same two peaks as described above, but it could now be seen that there was a shift in their locations, for quantiles 31–63% the peaks shifted to 26.1 °C and 77.1 °C. Archaea (Fig. 7(B,E)) had a single peak above 61% at 78.4 °C, and two peaks below at 32.9 °C and 93.9 °C. The Eukarya lacked peaks entirely (Fig. 7(C,F)).

We conducted recursive partitioning to find the best predictors of the association of strains with quantiles. For Bacteria, the highest taxonomic level associated was phylum; Proteobacteria, Firmicutes, Deinococcus-Thermus, and Aquificae were more often present in quantiles 40% and above, while Cyanobacteria, Actinobacteria, Bacteroidetes, Deferribacteres, Nitrospirae, and Planctomycetes below. Also in Bacteria, heterotrophs were more likely above the 30% quantiles and autotrophs below, and in the heterotrophs, facultative anaerobes were more often above the 60% quantiles. For Archaea and Eukarya no taxonomic, metabolic or trophic variables were significantly associated with quantiles.

It was apparent that there were groups of contiguous quantile curves in which the curves were highly parallel (Fig. 7). These groups were separated, one from the other, by discontinuities where the log-slopes of the curves abruptly shifted. This behaviour was quantified by plotting the log-slopes versus temperature, which showed horizontal lines separated by abrupt steps (results not shown). This parallelism was apparent in both ascending and descending curves in all three domains (Fig. 7), and in the Bacteria subgroups: pH and a_w tertiles, metabolic type and trophic type (Fig. 8).

3.2. Slope differences of quantile and growth curves

The differences in log-slopes of quantile and growth curves relative to U_{mes} are shown in Fig. 9. The credible intervals included

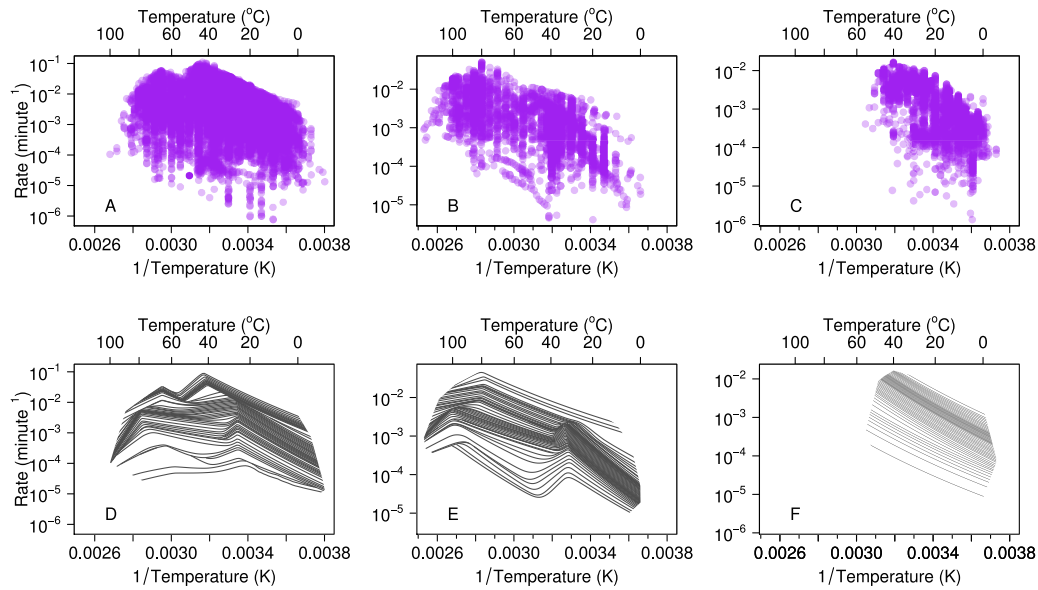


Fig. 7. The BKST plotted using logarithms of growth rate versus reciprocal absolute temperature by domain. The top row shows the observed growth rates using a logarithm scale versus reciprocal absolute temperature. The bottom row shows the fitted curves for quantiles 1%, 3%, 5%, ..., 99%. (A, D): Bacteria. (B, E): Archaea. (C, F): unicellular Eukarya.

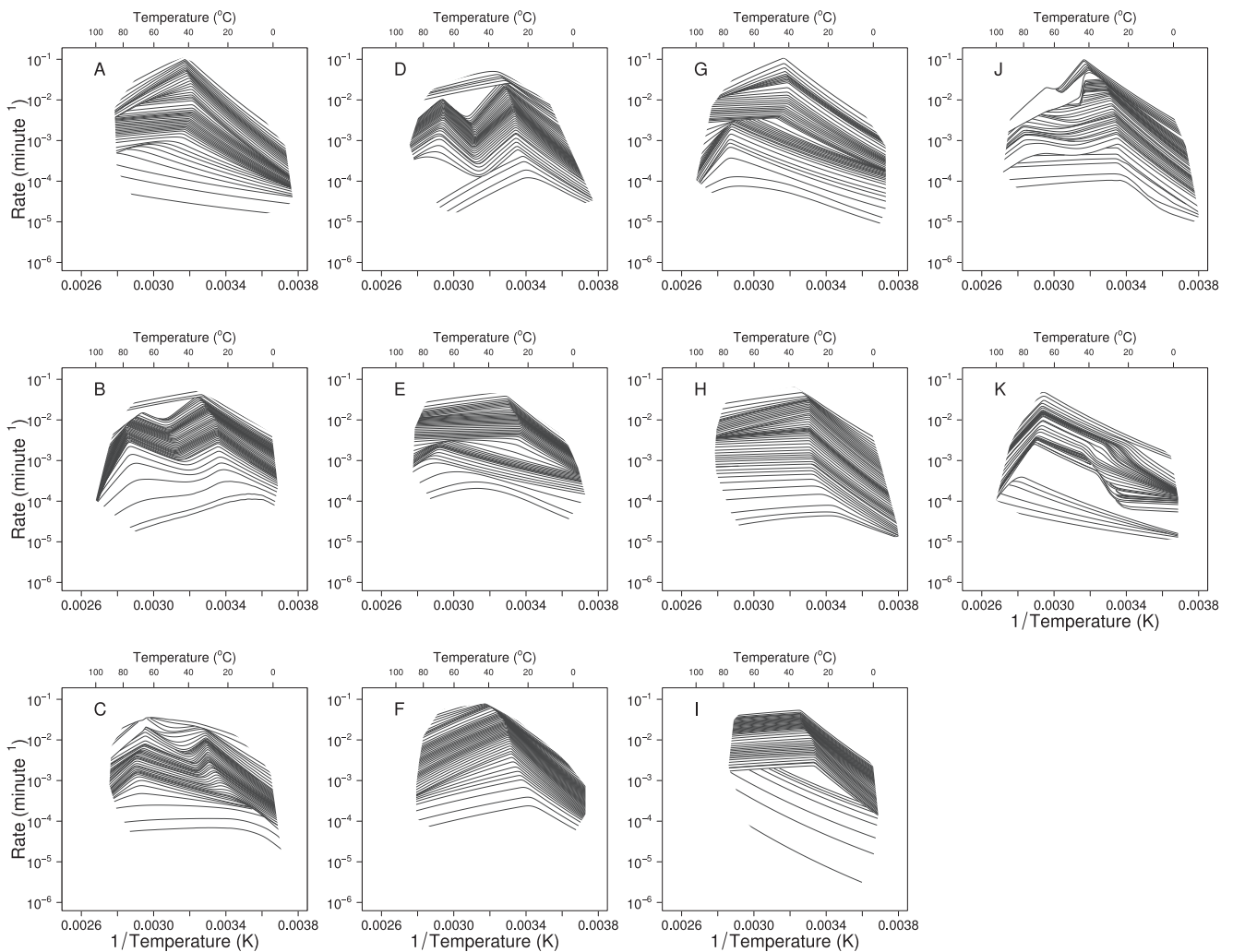


Fig. 8. The BKST for subgroups of Bacteria. Shown are quantile curves for: (A), lower pH tertile; (B), middle pH tertile; (C), upper pH tertile; (D), lower a_w tertile; (E), middle a_w tertile; (F), upper a_w tertile; (G), anaerobes; (H), aerobes; (I), facultative anaerobes; (J), heterotrophs; and (K), autotrophs.

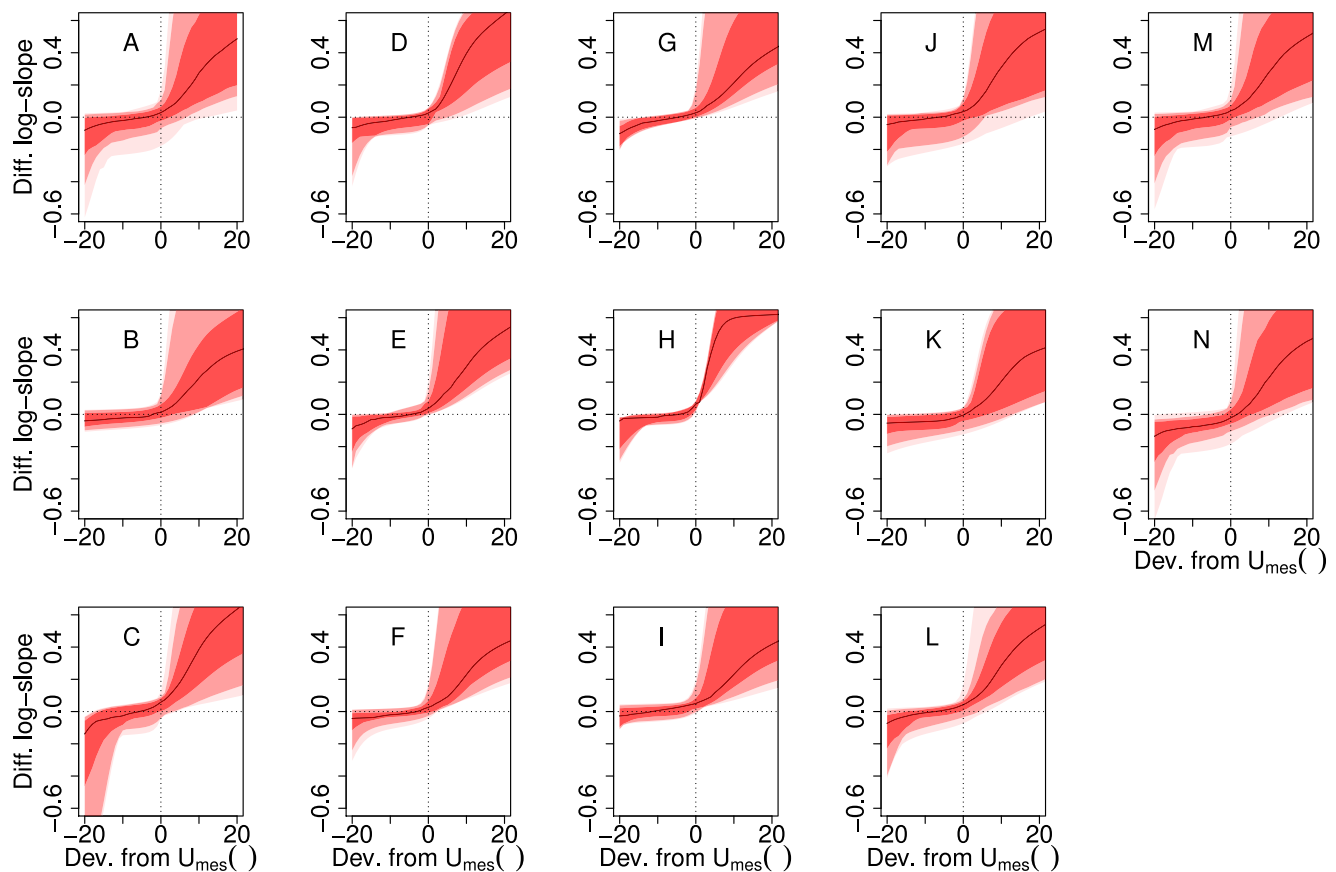


Fig. 9. Differences of log-slopes of fitted growth curves and quantile curves by domain (Bacteria, Archaea, Eukarya) and subgroups of Bacteria. Differences in log-slopes are plotted against temperatures relative to U_{mes} . Posterior means of the differences are represented by a solid line and the credible intervals shown as shaded bands, coloured dark red for 80%, medium red for 95%, and pale red for 99%. The domains were: (A), Bacteria; (B), Archaea; and (C), unicellular Eukarya. The subgroups were: (D), lower pH tertile; (E), middle pH tertile; (F), upper pH tertile; (G), lower a_w tertile; (H), middle a_w tertile; (I), upper a_w tertile; (J), anaerobes; (K), aerobes; (L), facultative anaerobes; (M), heterotrophs; and (N), autotrophs. a. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

a zero difference of slopes at U_{mes} for all domains and therefore the log-slopes of the quantile and growth curves at U_{mes} were the same with a 99% probability. Some credible intervals also included zero differences for negative deviations. In Bacteria there were also zero differences at U_{mes} for tertiles of pH and a_w , metabolic groups, and trophic type. This meant that the quantile curves had the same slopes as the growth curves at U_{mes} for all domains and subgroups with a 99% probability.

4. Discussion

We do not claim the assembled data set is a representative sample of life on Earth. The aim was to include as wide a range of strains as possible. We did not eliminate strains grown in suboptimal conditions and were more likely to include culturable strains of economic, veterinary, agricultural, or medical importance. A minority of strains in the data set were of varying physical sizes, but we did not concern ourselves with this, except to note that most of the data were microbial. In exploratory work we found using purely microbial data made no difference to the conclusions. Some may argue that little could be concluded from a disparate selection of strains, but nonetheless, there were temperature-dependencies found that were common to all strains.

4.1. The thermodynamic model

Biological growth rates depend on numerous processes and steps, but here we entertain the hypothesis that there is a single enzyme that is rate-limiting and common to all strains, which

would therefore have been conserved since the last universal common ancestor of the three domains. An alternative hypothesis is that growth is controlled by a complex network of enzymes. Dill et al. (2011) constructed a model in which denaturation of multiple proteins followed a statistical distribution, but we consider the fits of the growth rate curves so obtained insufficient. Such complex systems would also require feedback mechanisms to ensure physiological fluxes are maintained approximately constant over an extended temperature range (Ruoff et al., 2007), and involve many genes (Klumpp et al., 2009) that supervise complex transcriptomic and proteomic networks (Bruggeman and Westerhoff, 2007). If such complex genetic controls exist that would still not imply the absence of a single rate-limiting mechanism. Indeed, metabolic networks are probably identical for all living organisms (Jeong et al., 2000) raising the probability of a limiting mechanism. However, we do not exclude the possibility of a non-enzymic rate-limiting system, perhaps involving ribosomes (Fiedoruk et al., 2017), nor of a physical mechanism such as the role of water. Wiggins (2008) has suggested that water in close proximity to biomolecules can partition into high and low density states, thereby creating local osmotic pressure gradients, which in turn drives protein folding. The involvement of such mechanisms, if temperature-dependent, is not inconceivable.

We agree with those who argue that growth rate, all else being equal, can be a proxy for Darwinian fitness (Knies and King-solver, 2010) and hence be optimised at some particular temperature (Huey and Berrigan, 2001). This attribute represents a measure of performance that may be modified by natural selection,

and which is measurable and inheritable (Arnold, 1983). We might naively assume that this is T_{opt} , but this is the temperature of fastest growth and not a physiological optimum. According to the thermodynamic model at T_{opt} denaturation of the rate-limiting enzyme has increased sufficiently, and its activity concomitantly decreased, that it begins to offset the increase in growth rate that would otherwise occur in the absence of denaturation. Martin and Huey (2008) ingeniously proposed that an asymmetric growth curve in combination with symmetrically distributed environmental noise would result in Darwinian fitness being optimised at a temperature somewhere below T_{opt} . However, they did not provide a specific prediction for this optimum. A plausible candidate for an optimal temperature, and which does not require the presence of environmental noise, is U_{mes} , the temperature at which the cell grows as fast and also as efficiently as possible. The greater efficiency means that the cell can optimise its metabolism for growth with minimal demand on resources. We are unaware of other studies that have obtained a quantitative estimate of a temperature for optimal Darwinian fitness.

We heuristically defined U_{mes} to be the temperature at which the P_{NS} declines to 90% of its maximum, which approximates the upper shoulder of the P_{NS} curve. We acknowledge the arbitrariness, but in exploratory work we found that other cut points, such as T_{mes} , 95%, and 80%, resulted in less agreement in the slopes of the growth and quantile curves. Natural selection would be expected to trade-off growth rate and metabolic efficiency, and hence select a temperature above T_{mes} and below the stressful conditions of T_{opt} , which we think would be close to U_{mes} . We would also expect some variability around U_{mes} due to the exigencies of specific ecological conditions. U_{mes} can be quantified from any temperature-dependent growth curve to which the thermodynamic model can be applied, which represents an advance over less specific statements that fitness is optimised below T_{opt} (Martin and Huey, 2008).

The highest portions of the flat-topped P_{NS} curves were not perfectly flat, but possessed a very slight curvature in comparison to the rest of the curve (Fig. 3). We considered that this could be disregarded since natural selection would be expected to be more sensitive to the more highly curved than the less curved parts. In particular, fitness should be less sensitive to fluctuating environmental temperatures for strains with flat-topped P_{NS} curves compared to strains with peaked P_{NS} curves. This effect we would expect to be more obvious for higher temperatures if they are closer to the upper end of a flat-topped P_{NS} curve than towards its middle. This may explain why variability in environmental temperatures can reduce fitness at higher temperatures compared to lower temperatures (e.g., Bozinovic et al., 2011).

4.2. The Biokinetic Spectrum for Temperature

The BKST was described by us (Corkrey et al., 2016). In our initial report we described a data set of 1627 strains and 10,956 observations. Now, in this paper, with 2411 strains and 17,021 observations, we can see that the form of the BKST is largely unchanged (Fig. 2). The extents of the ranges of temperatures and growth rates in each are the same. The only visible modifications are an enhanced definition to the outline as the additional data further complete the spectrum. In this paper we estimate quantiles (1%, 3%, 5%, ..., 99%), whereas previously we only reported quantiles (50, 60, ..., 95, 97.5%). A more significant methodological difference is that in Corkrey et al. (2016) we only discussed the one-peak model. As shown in this paper, the no-peak and two-peak models have also proved useful since they have revealed biological differences between the domains. Whether the domain differences are the result of insufficient data remain unclear.

The most salient feature of the BKST was its roughly triangular appearance (Fig. 2). Visually examining the raw data, the upper boundary followed a curvilinear curve that began below 0 °C, reached a sharp peak at about 43 °C, before descending again to zero beyond 120 °C. The BKST spanned the entire temperature range over which growth rates have been reported, from −10 °C (Bakermans and Nealson, 2004) to 122 °C (Takai et al., 2008). It also included all three domains of life (Bacteria, Archaea and Eukarya) and the organism with the fastest known growth rate, *Clostridium perfringens*.

Organisms obviously use a variety of mechanisms to optimise growth rate under given conditions, but these adaptations do not invalidate the BKST. For example, psychrophiles need to overcome ecosystem hurdles such as nutrient availability, salinity (i.e., reduced water activity), freezing (ice crystal formation, desiccation, reduced water activity), and oxidative stress (O_2 supersaturation), and achieve this by using a variety of mechanisms, often in combination (Bowman, 2017). There are cellular traits closely connected to psychrophily, but few are exclusively connected to only cold adaptation since many have broad functionality (Bowman, 2017). Similarly, hyperthermophiles employ a wide range of strategies to compensate for the effects of elevated temperature such as increased protein stability (Kumar and Nussinov, 2001). Some organisms living in high salt concentrations have specific adaptations, such as the accumulation of concentrations of potassium, or the biosynthesis and/or accumulation of organic osmotic solutes ('compatible solutes', Oren, 2008). Thus, there are a multiplicity of adaptive traits that are modifications for particular conditions, whereas the BKST represents a fundamental limit to which all species conform.

The BKST also provides a reference to which the plausibility of models can be compared. Predictive growth rate models, such as are used in food science, ecology and climate change, must be consistent with the BKST; e.g., a model prediction found to be above the BKST upper boundary would be invalid. To put this idea on firmer ground requires a quantitative description of the BKST. To that end, we used quantile regression to fit models that were based on exponential curves (Eq. (3)). These had the merits of simplicity and symmetry. The prediction accuracies indicated that the models well described the data, though less well in the lower quantiles. Their form reflected the shape of the data distribution and since the ascending and descending curves were similarly parametrized their parameters had equivalent interpretations. In the BKST models, the B and D parameters controlled the slope and curvature of the quantile curve, A and C controlled the heights. If a large number of slower-growing species were to be appended to the existing data the A and C parameters would change their values and the quantile curves would shift upwards. But we expect that their log-slopes, set by the B and D parameters, would not change to any significant degree. We also expect that organism size would affect the A and C parameters and not the B and D parameters. This expectation is supported by the modest changes between Fig. 1 of Corkrey et al. (2016) and Fig. 2 shown herein.

We chose a model series of increasing complexity: the no-peak, one-peak, and two-peak models. This approach permitted a more detailed description of the data than earlier descriptions (Corkrey et al., 2016) by allowing each quantile to be separately described. In other words, the model used to describe a particular quantile curve was chosen independently of the other quantile curves. Selecting between models made use of a DIC, which tended to favour simpler models over more complex ones. It was also expected that where there were insufficient data, simpler models would be preferred, which may have resulted in the occasional switch in model types; e.g., in Archaea the one-peak model was preferred for the upper quantiles and the two-peak model for the lower quantiles (Fig. 7(E)). The two-peak model consisted of

two maxima, each with ascending and descending curves, with a trough separating them, which made it the most complex model. These procedures might conceivably have resulted in erratic assignments of models to quantile curves, but this proved not to be the case. Overall, the assignment of models to quantile curves displayed considerable order.

The locations of peaks and troughs suggested that all three domains were subject to common mechanisms. Peaks were often aligned within each domain and, while the peaks of Bacteria and Archaea differed from each other, there were also intriguing similarities. For example, both domains had a peak for quantiles below 63% that occurred at relatively similar temperatures (i.e., 26.1 °C and 32.3 °C). Assuming this was not the result of a sampling coincidence, it suggests that both domains are subject to similar limitations at those temperatures. Puurtinen et al. (2016) suggested that biomolecules are maximally stable in water at around 20 °C. It was also interesting that Bacteria and Archaea had troughs that were both located between 40 °C and 60 °C. The specific heat capacity of water (C_p) reaches a minimum at the slightly lower temperature of 36 °C (Dougherty and Howard, 1998). While this is interestingly close to the body temperatures of many endotherms, its closeness to the bacterial and archaeal troughs suggests a deeper connection. We may conjecture that instrumental in these phenomena is the global properties of water interacting with protein folding.

BKST substructure. It is well known that growth rates of organisms can vary by orders of magnitude and are temperature-dependent. This prompted the questions: are there limitations to the magnitude of growth rate and the shapes of growth curves? For the first part of the question, the upper boundary of the BKST corresponded to the maximum possible growth rate at any particular temperature, from which we made the *a posteriori* prediction that no form of life will be found to grow at a higher rate (Corkrey et al., 2018). For the second part, there was a limited range of log-slopes that could fit into the geometry of the BKST, hence, temperature dependency must also be limited. The existence of limits, as such, may be unsurprising or even intuitively expected, but if the cell's growth rate is actually a function of a complex metabolism then we might expect that these limits would have an equally complex form. The simplicity of the BKST should be surprising, even remarkable, and clearly requires an explanation.

In the BKST there were groups of contiguous, parallel quantile curves separated from adjacent groups by slightly different log-slopes. This phenomenon was observed in both the ascending and descending curves, but in this paper we are only concerned with the ascending curves in the BKST. Parallelism was unlikely to have resulted from sampling issues since it was apparent in all domains and other subgroups, such as for pH, a_w , metabolic and trophic types. As seen in Fig. 7 there appeared to be two major groups of parallel quantile curves, one for strains growing below the 63% quantile and the other for faster-growing strains above. A possible explanation was that the faster growing strains were similar to each other, perhaps taxonomically or ecologically, and hence shared the same temperature dependence, but differed in these respects from slower-growing strains. There were particular Bacteria phyla that were associated with lower and upper quantiles, and in the upper quantiles they were more likely to be heterotrophs with the fastest of them being facultative anaerobes. These associations may be linked to changes in log-slope between the groups of strains. But while we found these taxonomic and ecological groups in Bacteria, there were no associations in the other domains. This meant that, apart from the limits to growth rates and temperature-dependence already mentioned, we also had extensive groups of quantiles in a wide array of strains from disparate ecologies and taxonomies that had responded similarly. We concluded that par-

allelism was a global phenomenon that resulted from a common mechanism, influenced by taxonomy or ecological attributes, but which did not arise from them.

While a complex mechanism can produce simple results, we prefer to follow Occam's principle, opting for a less convoluted explanation over a more intricate one. The MRS can provide such a simple mechanism, though admittedly this is not a complete explanation since we have no candidate enzyme. The actual mechanism may be something other than a protein, a complex system of enzymes with intricately balanced feedback mechanisms, or a physical effect such as the influence of high and low density water. But the simpler hypothesis remains that the MRS is a rate-limiting enzyme, and it is this that results in the shape of the BKST and the quantile curves.

4.3. Slope differences of quantile and growth curves

Since the quantile curves were highly constrained, it followed that the growth rates constituting the BKST were also limited. The thermodynamic model well described such data and we expected it to share the same limitations. Therefore, we considered which growth curves intersected but did not exceed each quantile curve and at which temperature they were parallel. We had initially supposed that the contact point would be at T_{mes} , but T_{mes} was the centre of the NPR and that was essentially flat for many strains (q.v., Methods). Instead, we suspected the contact point would be at U_{mes} since at this temperature the cell should grow faster than at T_{mes} and almost as efficiently. The log-slopes of the growth curve and the nearest quantile curve were the same at U_{mes} in all domains (Bacteria, Archaea, and Eukarya) and Bacteria subgroups based on pH, a_w , metabolic and trophic types (Fig. 9). If there are multiple metabolic pathways that control growth then it may be conceivable that the ascending curves of the BKST would be exponential curves, but, if so, it would be difficult to see why their log-slopes should match that of the growth curves at this particular temperature. Therefore, this behaviour supported the MRS hypothesis.

The matching of log-slopes also favoured the Gamma hypothesis – that multiple inhibitory factors combine independently on growth (Bidlas and Lambert, 2008; Lambert and Bidlas, 2007a; 2007b). We might have expected non-zero differences with the more extreme pH or a_w , since the cell would presumably have to devote more resources to maintenance leaving less for reproduction as the conditions became more extreme, thereby modifying U_{mes} . This was not found to be the case. Since the mode of energy production in aerobic and anaerobic cells differ, we might have expected a difference in the shape of the growth curve resulting in a non-zero difference of log-slopes, but this also did not occur. Nor was a difference found for autotrophs and heterotrophs that are distinguished in how they obtain organic compounds. The BKST results indicate that U_{mes} is unaffected by any of these conditions. The current data set lacked data from simultaneous combinations of pH, a_w , or metabolic type on individual strains, preventing a direct test of the Gamma hypothesis. This would be of interest to confirm since it implies that the cell should perform optimally at an unvarying temperature. We do not claim that the growth rate does not change at U_{mes} when environmental conditions are changed (e.g., pH), only that the optimal temperature-dependency will be at this temperature.

U_{mes} can be seen as the temperature where an organism's Darwinian fitness is optimised, all else being equal, through a trade-off between growth rate and metabolic efficiency. Our results also showed that the quantile curves represent alternative combinations of temperature and growth rates possible for strains that have a particular log-slope at their respective U_{mes} . The quantile

curves, originally regarded by us as purely empirical (Corkrey et al., 2016), can now be seen to be the loci of growth rates and temperatures that optimise Darwinian fitness at a particular growth rate dependency.

4.4. Implications for evolutionary biology, ecology, and physiology

There are a number of results that bear on evolutionary biology, ecology, and physiology. We briefly outline these linkages here.

Evolutionary biology. We have shown that Darwinian fitness for a strain is maximised at the temperature U_{mes} . In a species that has a flat-topped P_{NS} curve, U_{mes} will be the upper limit of a potentially quite broad range of temperatures. This results from an enhanced stability of the rate-limiting protein. Psychrophiles and psychrotropes may additionally make use of compatible solutes by accumulating potassium, or by the biosynthesis and/or accumulation of organic osmotic solutes (Oren, 2008), thereby extending the range of temperatures over which they can grow. While compatible solutes are an important issue, we highlight here the role of the flat-topped P_{NS} curve in evolution.

Since U_{mes} is at the upper end of a potentially wide temperature range, random mutations in the rate-controlling protein are less likely to lead to reduced fitness. Most mutations are deleterious, but unless a random mutation in the rate-controlling enzyme changes the width of the P_{NS} curve it is not likely to affect the thermal stability of the organism between L_{mes} and U_{mes} . Controlling the rate of mutations comes at a physiological cost (Drake, 2009), but this would presumably be a reduced issue with proteins that are intrinsically more stable. Hence mutational costs will be lower in a species with a flat-topped P_{NS} curve than in a species with a peaked P_{NS} curve. A species with a flat-topped P_{NS} curve is also more likely to have higher evolutionary rates. This is because stable proteins are known to be more evolvable than other proteins since any random mutation is less likely to lead to loss of function (Bloom et al., 2006). In summary, strains with flat-topped P_{NS} curves will have higher evolutionary rates with lower mutational costs. It has been suggested that molecular clocks tick at rates determined by metabolic rates and temperature (Gillooly et al., 2005). Since temperature and mutation rates are correlated, then so are evolutionary rates with temperature. Therefore, at higher temperatures flat-topped P_{NS} curves are more advantageous. In support of this we have observed them to be more frequent in thermophiles (Corkrey et al., 2014). We expect that this would have been of considerable importance during the Precambrian if, as has been claimed (Knauth and Lowe, 1978), the early Earth was considerably warmer than today. Further, since temperature-dependence is likely to be an ecologically important characteristic of a strain, and since strains that fall along the same quantile curve have the same temperature-dependency, then random mutations will be more likely to result in evolutionary changes that track the quantile curve, rather than switching to a different curve. Hence, a random mutation is more likely to result in a shift along the quantile curves and not across them.

Ecology and climate change. Between L_{mes} and U_{mes} the organism is approximately equally efficient. In a species that has a peaked U_{mes} curve this will be a narrow range of temperatures, and in a species with a flat-topped U_{mes} curve it will be a wide range. Fitness is maximised at U_{mes} and will be reduced at T_{opt} . Of ecological interest is that there are species that have flat-topped P_{NS} curves. These species will be eurythermic since there will be no single temperature at which they perform poorly, though their fitness will still be maximised at U_{mes} . If these species are ecologically confined to a narrow temperature range, due to some isolating mechanism, then when the opportunity allows they will be able to cope with a wide range of temperatures. We have pre-

viously noted that flat-topped P_{NS} curves are more likely to be found in species with higher T_{opt} such as thermophiles and hyperthermophiles, but they also occur in psychrophiles (Corkrey et al., 2014). We posit that psychrophiles with peaked P_{NS} curves will be disadvantaged by climate change, but those with flat-topped P_{NS} curves may have the opportunity to extend their ranges. If a psychrophile has a flat-topped P_{NS} curve but has been historically confined in cold environments with temperatures below their U_{mes} , then it may be able to perform better if conditions change. This may have broad implications, especially since climate change is more likely to affect psychrophiles in complex ways (Vincent, 2010).

More generally, strains with peaked P_{NS} curves will be more vulnerable to random environmental fluctuations. For an individual strain, a rise in temperature beyond its U_{mes} would result in a decrease of its fitness. This is a qualitatively different situation to the observation sometimes made that the asymmetry of thermal profiles may render a species vulnerable to random fluctuations into the supraoptimal range (Chen and Shakhnovich, 2010). The strain may respond to a temperature rise by shifting along a quantile curve, thus maintaining the same temperature-dependence. This will be more easily accomplished by modifying phenotypically plastic attributes, such as the number of offspring, whereas a single rate-controlling enzyme is likely to be less easily modified due to its central role in the cell. As the strain shifts along the quantile curve it presumably maintains the same fitness (Brown et al., 2018). The concept is analogous to the ‘warmer is better’ hypothesis (Frazier et al., 2006; Huey and Kingsolver, 1989; Kingsolver and Huey, 2008), which is that selection tends to increase growth rate as the environmental temperature increases instead of keeping it fixed. Here, we provide a possible explanation for the phenomenon of why selection should favour faster growth rates as the temperature increases.

Physiology. Since quantile curves are the loci of growth rates and temperatures that optimise Darwinian fitness at a particular growth rate dependency, each includes species with relatively higher growth rates and higher T_{opt} , as well as relatively slower-growing organisms with lower T_{opt} . As suggested by Brown et al. (2018), smaller organisms with shorter lives and high productivity, and larger long lived organisms with lower productivity, can have approximately the same Darwinian fitness. Here, we show that organisms sharing the same quantile curve including those with relatively higher growth rates as well as relatively slower-growing organisms can all have the same temperature-dependence, and presumably the same fitness. From this, we can infer there should be phenotypic trade-offs with growth rate. In practice there may be several plausible alternative trade-offs, but we illustrate the idea with data on bacteria and other unicellular microbes from Lynch and Marinov (2015) who report that cell division times and cell volume are correlated. From their data we obtained the relationship $r = 0.00315 \cdot V^{-0.15}$ in which r is growth rate and V is cell volume. This indicates that growth rate declines as cell volume increases. This is a possible example of a trade-off between growth rate and soma production in organisms that may have the same temperature-dependence over a range of temperatures. This idea seems easily extended to multicellular organisms where trade-offs may exist between life-history traits affecting the soma and the germline (Berger et al., 2017).

5. Summary

The Biokinetic Spectrum for Temperature is a significant, and perhaps universal, biological phenomenon, and we believe it provides an insight into the nature of growth on Earth and elsewhere. To define the BKST quantitatively we used Bayesian quantile

regression and showed the result using quantile curves. The quantile curves displayed parallelism, suggesting temperature-dependent commonalities that spanned taxa and ecotypes, and suggested a common metabolic mechanism. The thermodynamic model posits a single rate-limiting enzyme common to all life that controls growth rate. The model predicted that there is a temperature, U_{mes} , at which the cell grows as fast and also as efficiently as possible. Darwinian fitness is likely to be optimised at U_{mes} , trading-off growth rate against physiological efficiency. This is a specific temperature that is, in principle, quantifiable from temperature-dependent growth curves. The fitness of a strain with a flat-topped P_{NS} curve, where the rate-controlling enzyme is stable over a wide range of temperatures, is expected to be more resilient to fluctuating environmental temperatures than a strain with a peaked P_{NS} curve, where the rate-controlling enzyme is only stable over a narrow temperature range.

The log-slopes of the ascending quantile curves were the same as the BKST growth curves at U_{mes} , which supported the MRS hypothesis. This was found for all domains and all subgroups of Bacteria, implying that optimal temperature-dependence occurs at U_{mes} and does not vary under the influence of other conditions such as pH, a_w , metabolic type and trophic type. Since this was the temperature at which fitness is optimised, U_{mes} , the ascending quantile curves in the BKST were the loci of the temperatures and growth rates that optimised fitness for a particular temperature-dependency. Our results indicate that, while growth rate and the viable temperature ranges of growth are limited to the extents of the BKST, there is only one optimal temperature-dependency for growth at each combination of growth rate and temperature. This may constitute an alternative explanation for why estimates of activation energies of growth rates from diverse species are often invariant (Brown et al., 2004).

Strains with flat-topped P_{NS} curves will be more evolvable, have higher random mutation rates, with lower mutational costs. Strains that fall along the same quantile curve will have the same temperature-dependence independently of other conditions such as pH, a_w , metabolic type and trophic type. In ecology, strains that have flat-topped P_{NS} curves are more likely to be eurythermic and, if they have been historically confined to cool conditions below their U_{mes} they will be able to perform better if conditions change. Adaptation is most likely to be achieved by shifting along a quantile curve, thus maintaining the same temperature-dependence and possibly the same fitness. Such changes will involve trade-offs by modifying phenotypically plastic attributes.

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Declarations of interest

none.

Supplementary material

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.jtbi.2018.10.052.

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